

Some Inorganic Constituents of the Muscles and Blood of the Oceanic Skipjack, *Katsuwonus pelamis*¹

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CELLULAR METABOLISM, anabolic and catabolic, regulates the production of macromolecules and the content of intracellular and extracellular ions. It is mainly the latter which are concerned with providing osmotic homeostasis. Much literature exists pertaining to the ionic composition of various tissues of aquatic and marine fish (Vinogradov, 1953). However, as noted by Love (1957), many of the values should be re-examined because of heterogeneity in sampling which makes comparisons almost impossible. Also, very little is known about the inorganic composition of a true pelagic form.

Only recently has it been possible to impound successfully for a fairly long period a fast swimming fish such as the oceanic skipjack, thus greatly enhancing the sampling of fresh tissues. Many of the fast-swimming pelagic fish have two distinct striated muscle types, white and red ("chiai"). When the tuna is cross-sectioned just anterior to the secondary dorsal fin, the two muscle types are readily discernible. The less plentiful red muscle pairs are located as bundles adjacent to the vertebrae. Both the dorsal and ventral pairs have horns that course, mediolaterally, toward the lateral line. At the skin the terminations of these horns are quite narrow. The remainder of the musculature is composed of the white muscle which approximately surrounds the red muscle bundles except at the lateral line.

Until recently, little was known about the physiological function of these two muscle types. It was thought that some indication might be obtained by determining the major

electrolyte composition of the muscles and of the blood. Also, the inorganic components of the oceanic skipjack, *Katsuwonus pelamis*, may be very informative to the comparative physiologist as well as to the nutritionist.

MATERIALS AND METHODS

The tuna ("aku") were caught from a ship by the barbless hook-pole fishing method in waters adjacent to Oahu, Hawaii. The fish were transported to Oahu in large circular tanks with a constant supply of circulating sea water. The tuna were impounded in tanks similar to, but larger than, those aboard ship. The fish were fed frozen smelt and beef liver once a day, and were sacrificed for sampling 24 hours after feeding.

Cardiac blood was drawn into heparinized syringes. After the hematocrit was determined, the plasma was prepared. The pH of the plasma was determined by using the Beckman Model G pH-meter fitted with microelectrodes. The plasma's osmolality was ascertained employing the Fiske Osmometer.

The carcasses were sectioned, after which pieces (1.5–2.0 gm) of the two muscle types were removed. After weighing, one-half of the samples were dried overnight at 110°C and the percentage water content was determined. These samples were digested in concentrated nitric acid, appropriately diluted, and the sodium and potassium contents determined. The remaining samples were homogenized in 10% trichloroacetic acid (TCA), centrifuged, and the supernatants were drawn off for the calcium, magnesium, and chloride analyses.

The sodium and potassium content of the plasma and the muscles were determined by flame spectrophotometry employing a Coleman Jr. spectrophotometer equipped with a propane-oxygen burner. The plasma calcium was ascertained by the Ferro-Ham method (Ferro and Ham, 1957a and 1957b), using a Beckman

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Model B spectrophotometer. The Geyer and Bowie (1961) method was employed to determine the calcium content of the muscles. Both the plasma and muscle magnesium were estimated by the flame spectrophotometric method of Van Fossan et al. (1959). An organic diluting solution was substituted for acetone. The composition of the former was 500 ml isopropanol, 74.6 mg KCl, 292 mg NaCl, and 300 ml deionized water. The muscle calcium and magnesium and the plasma magnesium samples were determined on a Beckman Model DU spectrophotometer equipped with a hydrogen-oxygen burner and a photomultiplier. The muscle and plasma chloride contents were determined using a Buchler-Cotlove chloridometer.

To ascertain whether there was a difference in the electrolyte composition of the red and white muscles, the data were statistically analyzed employing the standard t-test for group comparisons.

To compare the muscle results with those of other investigators, the inulin space of the two muscle types was determined using C^{14} -inulin. The tissues (about 50 mg) were incubated *in vitro* at 20°C in tuna physiological saline containing 2900 cpm/ml of inulin. The

composition of the saline was 19.99 mEq NaCl/liter, 7.52 mEq KCl/liter, 3.54 mEq $CaCl_2$ /liter, 2.70 mEq $MgSO_4$ /liter, 20 ml of phosphate buffer (pH = 7.4), and 4% glucose. At $\frac{1}{2}$, 1, 2, and 3 hours triplicate samples were removed from the media. After digestion in warm 30% KOH, 0.5-ml aliquots were transferred to pyrex planchettes and radioassayed on a Nuclear-Chicago Model 181 Scaler fitted with a DS-3 gas flow detector. All samples were corrected for self-absorption.

RESULTS

The data for the whole blood and plasma are contained in Table 1. The concentrations of the electrolytes and the osmolality are expressed in mEq/liter and mOs/liter, respectively. The bottom values under each column are the means \pm the standard error of the means (S.E.).

The mean values for the blood components were as follows: hematocrit = 52.61%, pH = 7.32, osmolality = 414.56, sodium = 203.74, potassium = 6.81, chloride = 177.43, calcium = 7.56, and magnesium = 2.22.

The electrolyte data for the red and white

TABLE 1
BLOOD COMPOSITION OF *K. pelamis*

| TUNA | HEMAT. | pH | OSM. ¹ | Na ² | K ² | Cl ² | Ca ² | Mg ² |
|------|--------|-------|-------------------|-----------------|----------------|-----------------|-----------------|-----------------|
| 1 | 56.0 | 7.50 | 450 | 200.3 | 4.70 | 178.00 | 7.81 | 3.35 |
| 2 | 54.0 | 7.18 | 416 | 196.2 | 6.78 | 189.67 | 7.92 | 2.27 |
| 3 | 45.0 | 7.38 | 400 | 194.9 | 5.71 | 186.67 | 6.54 | 1.66 |
| 4 | 48.0 | 7.30 | 396 | 185.5 | 6.56 | 176.00 | 7.14 | 1.30 |
| 5 | 51.0 | 7.29 | 394 | 186.2 | 9.64 | 171.00 | 7.46 | 1.80 |
| 6* | 58.0 | 7.25 | 428 | — | — | 162.00 | 10.55 | 2.68 |
| 7* | 52.0 | 7.38 | 445 | 231.8 | 2.81 | 189.00 | 7.08 | 4.05 |
| 8 | 48.0 | 7.41 | 400 | 200.0 | 7.50 | 176.00 | 7.04 | 1.55 |
| 9 | 39.0 | 7.33 | 401 | 200.5 | 5.97 | 176.00 | 6.28 | 1.53 |
| 10* | 59.0 | 7.41 | 436 | 222.7 | 3.85 | 182.00 | 9.41 | 3.18 |
| 11* | 59.0 | 7.42 | 425 | 213.6 | 8.85 | 181.00 | 7.64 | 2.32 |
| 12* | 55.0 | 7.22 | 426 | 211.4 | 8.71 | 177.00 | 8.37 | 2.70 |
| 13 | 52.0 | 7.31 | 406 | 200.0 | 11.60 | 176.50 | 6.51 | 1.45 |
| 14 | 53.0 | 7.22 | 416 | 202.5 | 8.10 | 175.00 | 7.43 | 2.35 |
| 15 | 51.0 | 7.18 | 414 | 209.5 | 3.80 | 174.50 | 7.46 | 1.72 |
| 16 | 46.0 | — | 397 | 201.0 | 7.60 | 173.00 | 6.19 | 1.59 |
| 17* | 57.0 | — | 398 | — | — | 173.00 | 7.22 | 2.00 |
| 18 | 64.0 | — | 414 | — | — | — | 8.13 | 2.41 |
| Mean | 52.61 | 7.32 | 414.56 | 203.74 | 6.81 | 177.43 | 7.56 | 2.22 |
| S.E. | 1.417 | 0.025 | 4.082 | 3.216 | 0.624 | 1.652 | 0.256 | 0.177 |

* Hemolyzed blood.

¹ Osmolality values in mOs/liter.

² Electrolyte values in mEq/liter.

TABLE 2
RED MUSCLE COMPOSITION OF *K. pelamis*¹

| TUNA | % H ₂ O | Na | K | Cl | Ca | Mg |
|------|--------------------|-------|-------|-------|-------|-------|
| 1 | 70.99 | 26.50 | 75.70 | — | — | 11.80 |
| 2 | — | 22.12 | 75.32 | 0.0 | 0.00 | 18.43 |
| 3 | — | 26.94 | 74.56 | 0.0 | 0.86 | 20.26 |
| 4 | 72.83 | 29.14 | 80.40 | — | 0.51 | 8.49 |
| 5 | 75.64 | 22.62 | 73.35 | — | 9.11 | 11.72 |
| 6 | 72.49 | 21.32 | 92.65 | — | 3.04 | 21.98 |
| 7 | 72.60 | 15.80 | 91.80 | 15.96 | 0.66 | 21.59 |
| 8 | 72.03 | 18.36 | 76.26 | — | 1.56 | 19.44 |
| 9 | 75.09 | 26.53 | 78.66 | 52.17 | 1.07 | 18.24 |
| 10 | 72.90 | 20.30 | 94.40 | 42.14 | — | — |
| 11 | 72.31 | 18.74 | 94.40 | 61.74 | 0.93 | 21.29 |
| 12 | 72.08 | 25.18 | 78.06 | — | 1.40 | 18.43 |
| 13 | 73.53 | 14.56 | 69.86 | — | 1.45 | 19.06 |
| 14 | 72.16 | 16.93 | 75.83 | 38.74 | 1.22 | 16.95 |
| 15 | 72.18 | 14.63 | 71.80 | 28.10 | 1.78 | 17.09 |
| 16 | 73.46 | 20.30 | 80.40 | 67.03 | 4.70 | 14.70 |
| 17 | 73.42 | 18.40 | 84.40 | 55.58 | 0.78 | 14.51 |
| 18 | 70.79 | 18.10 | 80.10 | 73.66 | 0.74 | 16.02 |
| Mean | 72.78 | 20.92 | 80.44 | 39.56 | 1.86 | 17.06 |
| S.E. | 0.318 | 1.050 | 1.853 | 7.48 | 0.557 | 0.920 |

¹ Electrolyte values in mEq/kg fresh wt.

TABLE 3
WHITE MUSCLE COMPOSITION OF *K. pelamis*¹

| TUNA | % H ₂ O | Na | K | Cl | Ca | Mg |
|------|--------------------|-------|--------|-------|-------|-------|
| 1 | 71.75 | 16.20 | 94.60 | — | 58.80 | 19.70 |
| 2 | — | 9.12 | 98.79 | 23.58 | 7.12 | 27.29 |
| 3 | — | 17.16 | 109.38 | — | 5.00 | 26.38 |
| 4 | 76.91 | 17.44 | 113.52 | — | 0.94 | 17.13 |
| 5 | 80.24 | 22.41 | 113.44 | — | 0.44 | 16.49 |
| 6 | 72.13 | 7.26 | 88.35 | 0.00 | 3.99 | 33.79 |
| 7 | 71.34 | 9.23 | 113.35 | 0.00 | 0.52 | 31.71 |
| 8 | 73.97 | 9.20 | 116.70 | 0.00 | 1.48 | 25.49 |
| 9 | 76.69 | 26.71 | 109.36 | 77.34 | 2.34 | 24.38 |
| 10 | 71.25 | 6.95 | 94.20 | 87.57 | 0.00 | 35.02 |
| 11 | 76.65 | 9.89 | 114.60 | 91.36 | 0.00 | 23.98 |
| 12 | 73.19 | 3.61 | 125.90 | 44.73 | 0.83 | 27.59 |
| 13 | 75.14 | 7.14 | 95.53 | 36.93 | 1.25 | 25.86 |
| 14 | 74.16 | 8.12 | 108.43 | 55.56 | 1.85 | 25.20 |
| 15 | 74.42 | 7.18 | 100.73 | 52.80 | 0.93 | 22.78 |
| 16 | 75.44 | 11.50 | 108.50 | 20.30 | 2.86 | 20.42 |
| 17 | 75.28 | 8.70 | 105.00 | 16.23 | 0.18 | 18.95 |
| 18 | 73.29 | 9.70 | 104.40 | 19.92 | 0.27 | 21.90 |
| Mean | 74.49 | 11.53 | 106.38 | 37.59 | 4.93 | 24.67 |
| S.E. | 0.603 | 1.422 | 2.255 | 8.480 | 3.200 | 1.241 |

¹ Electrolyte values in mEq/kg fresh wt.

muscles comprise Tables 2 and 3, respectively. As in the former table, the last values are the means \pm the S.E. The values in these tables, excluding the water content, are in mEq/kg

fresh weight. A brief resume of the statistical comparisons is contained in Table 4.

The inorganic contents of the red and white muscles, respectively, were: water = 72.78,

TABLE 4
VALUES OF T-TESTS FROM DATA IN TABLES 2 AND 3.

| ELECT. | MUSCLE | MEAN (mEq/kg) | MEAN ₁ -MEAN ₂ | CALCULATED t-VALUE | P VALUE |
|--------------------|--------|------------------|--------------------------------------|-----------------------|---------|
| % H ₂ O | red | 72.78 | 2.0 | 2.947* | 0.01 |
| | white | 74.49 | | | |
| Na ⁺ | red | 20.92 | 9.39 | 5.314* | 0.001 |
| | white | 11.53 | | | |
| K ⁺ | red | 80.44 | 25.94 | 8.890* | 0.001 |
| | white | 106.38 | | | |
| Ca ⁺⁺ | red | 1.86 | 3.07 | 0.892 | 0.40 |
| | white | 4.93 | | | |
| Mg ⁺⁺ | red | 17.06 | 7.61 | 4.744* | 0.001 |
| | white | 24.67 | | | |
| Cl ⁻ | red | 92.16 | 23.40 | 0.549 | 0.50 |
| | white | 68.76 | | | |

* Significant difference.

74.49%; sodium = 20.92, 11.53; potassium = 80.44, 106.38; calcium = 1.86, 4.93; magnesium = 17.06, 24.67; and chloride = 92.16, 68.76.

Statistical differences between the means demonstrated that only the calcium and chloride contents of the two muscle types were not significantly different.

The uptake of the C¹⁴-inulin by the two muscle types is illustrated in Figure 1. The two intercepts at the ordinate were fitted by eye. The values for the red and white muscles are 0.69 and 0.55 cpm/mg fresh weight, respectively. The calculated extracellular (inulin) space for the red muscle is 23.79% and that of the white muscle is 18.97%.

Table 5 contains the comparison of some plasma constituents of various fishes with those of the skipjack, *K. pelamis*. The plasma concentrations are in mEq/liter.

The comparison of the muscle electrolyte contents of some teleosts with those found in the skipjack is contained in Table 6. The concentrations are expressed in mEq/kg tissue water.

DISCUSSION AND CONCLUSIONS

The blood constituents of *K. pelamis* (Table 1) will be discussed with comparable data from other species in a later section of this presentation.

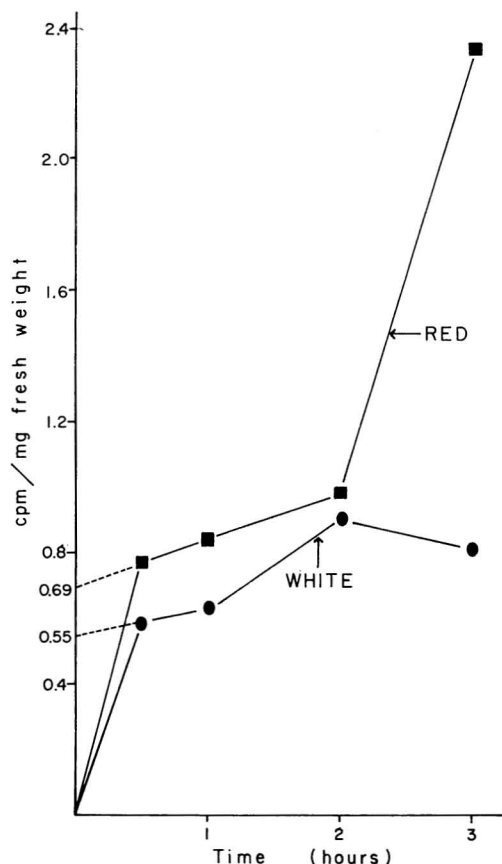


FIG. 1. Uptake, *in vitro*, of C¹⁴-inulin by red and white muscles of the skipjack, *K. pelamis*.

TABLE 5
COMPARISON OF SOME PLASMA CONSTITUENTS OF VARIOUS MARINE FISHES

| FISH | Na ¹ | K ¹ | Cl ¹ | Ca ¹ | Mg ¹ | OSM. ² | REFERENCE |
|---|-----------------|----------------|-----------------|-----------------|-----------------|-------------------|----------------------------------|
| Cyclostomata | | | | | | | |
| <i>Polistotrema stoutii</i> (hagfish) | 544 | 7.7 | 446 | 10.8 | 20.8 | | Urist (1963) |
| <i>Myxine glutinosa</i> (hagfish) | 558 | 9.6 | 576 | 12.4 | 38.8 | | Robertson (1960) |
| Chondrichthyes | | | | | | | |
| <i>Raja erinacea</i> (skate) | 254 | 8 | 225 | 12 | 5 | 1035* | Hartman et al. (1941) |
| <i>Narcine</i> sp. | 134 | 7 | 159 | 24 | 6 | | Salome Pereira and Sawaya (1957) |
| <i>Rhinobatus</i> sp. | 143 | 12.8 | 143.6 | 14.6 | 4 | | Salome Pereira and Sawaya (1957) |
| <i>Triakus semifasciatus</i> (shark) | 235 | 10 | 230 | 10 | 6 | | Urist (1962) |
| Teleostei | | | | | | | |
| <i>Muraena ? atlanticus</i> (eel) | 101 | 6.2 | 140 | 5.0 | 2.4 | | Urist (1963) |
| <i>Muraena helena</i> (eel) | 212 | 1.95 | 188 | 7.74 | 4.84 | | Robertson (1960) |
| <i>Lophius piscatorius</i> (goosefish) | 193 | 5 | 153 | 12 | 10 | 431* | Brull and Cuypers (1955) |
| <i>Lophius piscatorius</i> (goosefish) | 185 | 5.1 | 153 | 6.4 | 5.0 | | Robertson (1954) |
| <i>Paralabrax clathratus</i> (kelp bass) | 180 | 5.0 | 147 | 6.0 | 3.0 | | Urist (1962) |
| <i>Scomberomorus maculatus</i> (mackerel) | 198 | 10.3 | 176 | | | | Becker et al. (1958) |
| <i>Clupea harengus</i> (herring) | | | | | | 512 | Holliday and Blaxter (1961) |
| (barracuda) | 215 | 6.4 | 189 | | | | Becker et al. (1958) |
| <i>Mycteroperca bonaci</i> (grouper) | 237 | 8.2 | 217 | | | | Becker et al. (1958) |
| <i>Katsuwonus pelamis</i> (skipjack) | 204 | 6.8 | 177 | 7.6 | 2.2 | 415 | authors (1966) |

¹ Electrolyte values in mEq/liter.

² Osmolality values in mOs/liter.

* From Potts and Parry (1964).

The data contained in Tables 2 and 3 were analyzed statistically to determine whether the concentrations differed with muscle type; these results are presented in Table 4. The white muscle contains a greater amount of water, potassium, and magnesium which possibly indicates that this tissue has a larger intracellular space. Statistical differences in the calcium and chloride contents were not found, which may have been due to the TCA used in the extraction. However, blanks were carried throughout the analysis.

Figure 1 illustrates that the extracellular volume of the red muscles is greater than that of the white. The calculated inulin spaces for the red and white muscles are 23.79 and 18.97%, respectively. The higher sodium content of the red muscle verifies the larger extracellular space.

It was not possible to determine accurately the intracellular sodium and chloride contents because of their high content in the intravascular compartment. The other intracellular calculated concentrations (mEq/liter cell water) of the red and white muscles, respectively, were: potassium = 160.88, 189.28; magnesium = 33.76, 43.68; calcium = 0.12, 6.30.

It is well known that the amount of intracellular potassium determines the threshold value for any tissue. Thus, the potential produced by this ion for the red muscle was calculated to be 83.77 mv and that of the white muscle was 88.08 mv—a potential difference of 4.31 mv. Therefore, the red muscle would have a lower threshold value, indicating that possibly this muscle would be utilized more than the white.

Vernick (1964) reported that the red muscle of four pelagic species had a higher content of thiamine, riboflavin, pantothenic acid, vitamin B₁₂, myoglobin, and cytochrome C. This tissue also had a higher degree of vascularization and larger mitochondria in the sarcoplasm. These findings led to the suggestion that the red muscle provided energy for the white. Hamamoto and Hohl (personal communication) discovered that the mitochondrial density in the red muscle sarcoplasm of *K. pelamis* was approximately one magnitude greater than that in the white. Because the mitochondria are the cell's energy producers, there is a strong correlation between the degree of activity of

the muscle and the number and shape of the mitochondria within the muscle cells (Davson, 1964). In addition, if one considers the color of the two muscle types and applies the analogy of the breast muscles of chickens versus those of the pigeon, it becomes apparent that the red muscle of *K. pelamis* with its abundance of mitochondria is possibly used for swimming and not as an energy producer for the white muscle. The red muscle is, indeed, able to contract and is probably used for normal swimming activity (Rayner, personal communication). The white muscle may be used secondarily, e.g., for accelerated and rapid movements seen during avoidance and feeding reactions.

Table 5 lists some of the plasma constituents of various fishes. It is well known that the marine cyclostomes are approximately isosmotic to the medium and that the marine cartilaginous fishes are hyperosmotic to the environment. However, the sea water-inhabiting teleosts are hyposmotic to their medium. Thus, these animals are threatened by desiccation. To prevent dehydration the animals must drink water and selectively excrete ions. The latter process is generally accomplished extrarenally via the gills.

Of the teleosts listed in Tables 5 and 6 only the barracuda and herring can be comparable to the skipjack, and the eels would be intermediate in comparison; the other species would be least comparable due to their phylogenetic placement and their relative inactivity as compared with the scombroid fishes. The mackerel is a scombroid fish, but it inhabits more inshore waters than does the skipjack.

As expected, the electrolyte composition of the skipjack plasma (Table 5) is less than that of the cyclostomes. However, it approximates those of the chondrichthyes. The greater osmolality of the latter is due to a higher urea content of the plasma. Concentrations of 300–400 mM of urea and trimethylamine oxide/liter are essential for elasmobranch osmoregulation (Urist, 1962). The plasma calcium and magnesium in the skipjack are much less than those in the chondrichthyes. This can be attributed to the apatite, which allows the teleost to maintain ionic concentrations independent of the external medium, and to the greater efficiency of the kidney and possibly the gills.

TABLE 6
COMPARISON OF SOME MUSCLE CONSTITUENTS OF SOME MARINE TELEOSTS¹

| FISH | Na | K | Cl | Ca | Mg | REFERENCE |
|---|-------|--------|-------|-------|-------|-------------------------|
| <i>Muraena helena</i> (eel) | 25.0 | 165 | 23.7 | 18.7 | 14.9 | Robertson (1960) |
| <i>Mycteroperca bonaci</i> (grouper) | 51.5 | 125.5 | 26.7 | | | Becker et al. (1958) |
| <i>Scomberomorus maculatus</i> (mackerel) | 71.7 | 153.5 | 53.8 | | | Becker et al. (1958) |
| <i>Clupea pilchardus</i> (herring) | 53.8 | 170.6 | 65.6 | 152.1 | 51.3 | Carteni and Aloj (1934) |
| <i>Katsuwonus pelamis</i> (skipjack) | | | | | | |
| Red muscle | 35.46 | 136.36 | 67.06 | 3.15 | 28.92 | authors (1967) |
| White muscle | 20.77 | 191.61 | 67.71 | 8.88 | 44.43 | |

¹ Values in mEq/kg water.

The plasma sodium content of the skipjack is greater than those of the Atlantic eel, the goosefish, and the kelp bass, but is lower than those of the Roman eel, the barracuda, and the grouper. The same order is found for the skipjack when the chloride values are compared. In comparing the potassium values, the Atlantic eel and the barracuda have similar concentrations of plasma chlorides. The Roman eel, the goosefish, and the kelp bass have lower plasma chlorides than the skipjack. In *K. pelamis*, lesser concentrations of plasma sodium, potassium, and chloride are probably due to differences in the osmoregulatory mechanism and the type of integument. Excluding the mackerel and possibly the barracuda, the listed teleosts are not true pelagic species and may be subjected to some degree of salinity fluctuations. In Hawaii, the barracuda is frequently seen in shallow lagoons which are subjected to dilutions during heavy rains.

Comparison of the plasma calcium of the listed teleosts shows that only the goosefish (Brull and Cuypers, 1955) had a higher plasma content. The value reported by Robertson (1954) appears to be in agreement with those reported by the other investigators. It is also apparent that the plasma magnesium of *Lophius* is greater than those reported by the other authors. It is known that temperature plays an important role in the solubility product constant of compounds and, thus, the rate of ionic exchange in apatite. A higher body temperature coupled with high serum alkaline phosphatase activity and other factors would favor a decrease in blood calcium, phosphate, and magnesium. It has been reported that tunas and skipjacks have body temperatures 6°–12°C

higher than their environment (Kishinouye, 1923; Berg, 1940; Morrow and Mauro, 1950; and Van Oosten, 1957). It would be expected, then, that the blood calcium and magnesium content of the skipjack would be less than that found in the colder poikilothermic fishes, but greater than that found in mammals. This is apparent for the magnesium values but not for those of the plasma calcium. Also, there is a correlation between the activity of species and the amount of plasma magnesium. The more active forms generally have lesser concentrations of plasma magnesium. The greater blood calcium level of the skipjack, excluding the value of Brull and Cuypers, may be due to the intrinsic factors controlling osteogenesis and the amount of apatite coupled with the efficiency of the kidney, the ionic strength of the serum, and the amount of vitamins A and D stored in the liver.

In brief, the differences in blood ionic concentrations of various fishes is greatly influenced by the type and composition of the skeleton. Apatite not only stores Ca^{+2} and PO_4^{-2} but also Na^+ , Mg^{+2} , and CO_3^{-2} . The regulation of K^+ and Cl^- is influenced not by the skeleton but by the gills and kidneys. Osmoregulation is delicately controlled by enzymes, hormones, and vitamins. The amounts and activities of these complexes are influenced by intrinsic and extrinsic factors which affect cell permeability and metabolism in such a way that each organism is unique in its electrolyte composition.

Comparative values of some muscle electrolytes of marine teleosts are presented in Table 6. The ionic composition of the plasma definitely influences that of the surrounding tissues.

Fish with high plasma electrolyte values usually have tissues with relatively high electrolyte values. The grouper, mackerel, and herring have greater amounts of muscle sodium than does the skipjack. The sodium content of the muscles of the eel is intermediate between the skipjack's red and white muscle content. This is to be expected because the plasma sodium of *K. pelamis* closely approximates that of the eel. However, it is also apparent that the blood sodium of the mackerel and the skipjack are present in nearly equal concentrations. At first glance the difference in the muscle sodium content of the two species is obscure, but it will be recalled that the body temperature of tuna is 6°–12°C higher than their environment and, therefore, the muscles of the skipjack would probably be more active metabolically than those of the mackerel. If this is truly the case, the sodium pump of the skipjack would be much more efficient, thus producing a lesser intracellular sodium content than that present in the mackerel and possibly in the other higher teleosts.

Upon comparing the potassium content of the various muscles, it becomes apparent that the plasma content does not necessarily influence the muscle content. This is obvious on examining the values for *Muraena* and *Scomberomorus*. The plasma potassium content of the former animal is 1.95 mEq/liter in contrast to a muscle content of 165 mEq/kg water. The potassium content of the plasma for *Scomberomorus* is 10.3 mEq/liter as compared with a muscle content of 153 mEq/kg water. Also, on examination of the blood and muscle concentration of the teleosts, no obvious order is evident, e.g., the blood potassium order is: mackerel > grouper > skipjack > eel, and the muscle order is: skipjack white > eel > mackerel > skipjack red > grouper. It appears, therefore, that the difference in muscle potassium may be under greater metabolic control than is sodium. Thus, the extracellular potassium may be entirely under the influence of the hormonal and genetic composition of the animal.

The chloride content of a tissue, like the sodium content, is greater extracellularly than intracellularly. It would then be expected that the chloride content of the muscles would parallel that of the plasma. However, the data in

Tables 5 and 6 do not support this hypothesis. The importance of chloride in a tissue is to maintain electrochemical neutrality. Thus, the chloride content of a tissue is maintained passively as a result of the Na⁺ and K⁺ distribution. As noted above, this ionic distribution is genetically influenced and thus the Cl⁻ distribution would subsequently be controlled but in a more subtle manner. Further examination of Table 6 reveals that the muscle and blood chlorides of the grouper are greater than those of the eel, and also that the chloride values for the skipjack are greater than those of the mackerel. However, the relationship between blood and muscle chlorides terminates at this point, because the skipjack muscles have the greatest chloride content, but the plasma chloride content is intermediate between those of the eel and the mackerel.

It is not possible to make similar comparisons with the herring because the blood values of this fish could not be located. Such data would be informative because the herring is more closely related systematically and ecologically to the skipjack than to the eel and grouper.

The data for muscle calcium and magnesium of marine teleosts are very meager. In Table 6 only one direct comparison can be made, that between *M. helena* and *K. pelamis*. The values for the herring cannot be considered because, as was noted by Robertson (1960), the muscle samples were contaminated with bone fragments. The calcium content of both muscle types of the skipjack is less than that of the eel, although the blood calcium levels of both species are approximately the same.

The results of the comparison of the magnesium contents are opposite to those of the calcium comparison. The eel has about twice the amount of blood magnesium that the skipjack does. The differences in the muscle content are that the red muscle of *K. pelamis* has about twice the amount, and the white muscle has about three times the amount found in the eel. This may be due to a greater preponderance of myosin and adenosine triphosphate (ATP) in the muscles of the skipjack.

It is known that magnesium serves as a cofactor for bridging ATP and creatine to the creatine kinase molecule during transphosphorylation (White et al., 1964). It is quite pos-

sible that the muscle ATP content of fast swimming fish is greater than that in less active forms. Studies on the phosphorus compounds of fish muscle may produce a strong correlation between magnesium and ATP-creatine phosphate contents.

SUMMARY

1. The major electrolyte constituents of the plasma, red muscle, and white muscle of the oceanic skipjack, *Katsuwonus pelamis*, were determined. The potassium content and the greater mitochondrial density of the red muscle suggest that this muscle is utilized for normal swimming activity rather than being an energy source for the white muscle.

2. The plasma electrolytes were compared with those of other marine fishes. In general, the sodium content of the skipjack plasma is less than that found in the cyclostomes, the skate and the shark, but is slightly greater than that found in the majority of other teleosts. The plasma potassium is less than that in the cyclostomes and elasmobranchs and greater than that in other teleosts. The plasma chloride content of the skipjack, as well as the calcium and magnesium, is less than that of the other investigated species.

3. Comparison of the differences in the electrolyte composition of the red and white muscles reveals that the white tissue contains a larger amount of water, potassium, and magnesium. However, the red muscle contains a greater amount of sodium.

4. Using C^{14} -inulin, the extracellular space of the red and white muscles was determined to be approximately 0.24 l/kg muscle and 0.19 l/kg muscle, respectively.

5. The muscle electrolyte content of *K. pelamis* was contrasted with the muscle contents of other teleosts. The order of decreasing composition is as follows. For Na^+ : mackerel > herring > grouper > skipjack red > eel > skipjack white; for K^+ : skipjack white > herring > eel > mackerel > skipjack red > grouper; for Cl^- : skipjack red > skipjack white > herring > mackerel > grouper > eel. Both muscle types of the skipjack contained less calcium and more magnesium than did the muscle of the eel.

REFERENCES

- BECKER, E. L., R. BIRD, J. W. KELLY, S. S. SCHILLING, and N. YOUNG. 1958. Physiology of marine teleosts. I. Ionic composition of the tissue. *Physiol. Zool.* 31:224.
- BERG, L. S. 1940. Classification of fishes both recent and fossil. *Trav. Inst. Zool. Acad. Sci. USSR* 5:517.
- BRULL, L., and Y. CUYPERS. 1955. Blood perfusion of the kidney of *Lophius piscatorius* L. IV. Magnesium excretion. *J. Mar. Biol. Assn. U. K.* 34:637.
- CARTENI, A., and G. ALOJ. 1934. Composizione chimica de animali marini del golfo de Napoli. I. Teleostei. *Arch. Internat. Physiol.* 42:398.
- DAVSON, H. 1964. A Textbook of General Physiology. 3rd ed. Little, Brown and Co., Boston. 1166 pp.
- FERRO, P. V., and A. M. HAM. 1957a. A simple spectrophotometric method for the determination of calcium. *Am. J. Clin. Path.* 28:208.
- 1957b. A simple spectrophotometric method for the determination of calcium. II. A semimicro method with reduced precipitation time. *Am. J. Clin. Path.* 28:689.
- GEYER, R. P., and E. J. BOWIE. 1961. The direct determination of tissue calcium by flame photometry. *Anal. Biochem.* 2:360.
- HARTMAN, F. A., L. A. LEWIS, K. A. BROWNELL, F. F. SHELDON, and R. F. WALTHER. 1941. Some blood constituents of the normal skate. *Physiol. Zool.* 14:476.
- C. A. ANGERER, and F. A. SHELDON. 1944. Effect of interrenalec-tomy on some blood constituents in the skate. *Physiol. Zool.* 17:228.
- HOLLIDAY, F. G. T., and J. H. S. BLAXTER. 1961. The effects of salinity on the herring after metamorphosis. *J. Mar. Biol. Assn. U. K.* 41:37.
- KISHINOUE, K. 1923. Contributions to the comparative study of the so-called scombroid fishes. *J. Coll. Agr. Imp. Univ., Tokyo* 8:293.
- LOVE, R. M. 1957. The biochemical composition of fish, Chapt. 10, pp. 401-418. In: M. E. Brown, ed., *The Physiology of Fishes*, Vol. 1. Academic Press, N. Y.

- MORROW, J. E., JR., and A. MAURO. 1950. Body temperatures of some marine fishes. *Copeia* 2:108.
- POTTS, W. T. W., and G. PARRY. 1964. Osmotic and Ionic Regulation in Animals. Macmillan Co., N.Y. 423 pp.
- ROBERTSON, J. D. 1954. The chemical composition of the blood of some aquatic chordates, including members of the Tunicata, Cyclostomata and Osteichthyes. *J. Exptl. Biol.* 31:424.
- . 1960. Studies on the chemical composition of muscle tissue. I. The muscle of the hagfish, *Myxine glutinosa*, and the Roman eel, *Muraena helena*. *J. Exptl. Biol.* 37:879.
- SALOME PEREIRA, R., and P. SAWAYA. 1957. Ions in the bloods of elasmobranchs. *Bol. Fac. Fil. Cien Univ., São Paulo, Zool.* 21.
- SMITH, H. W. 1931. The absorption and excretion of water and salts by the elasmobranch fishes. II. Marine elasmobranchs. *Am. J. Physiol.* 98:269.
- URIST, M. R. 1962. The bone-body fluid continuum: calcium and phosphorus in the skeleton and blood of extinct and living vertebrates. *Perspect. Biol. Med.* 6:75.
- . 1963. The regulation of calcium and other ions in the serums of hagfish and lampreys. *Ann. N.Y. Acad. Sci.* 109:294.
- VAN FOSSAN, D. D., E. E. BAIRD, and G. S. TEKELL. 1959. A simplified flame spectrophotometric method for estimation of magnesium in serum. *Am. J. Clin. Path.* 31:368.
- VAN OOSTEN, J. 1957. The skin and scales, Chapt. 5, pp. 207–244. In: M. E. Brown, ed., *The Physiology of Fishes*, Vol. 1. Academic Press, N.Y.
- VERNICK, S. H. 1964. Histology of the red muscle in four teleosts. *Copeia* 4 (44th Ann. Meet. Am. Soc. Ichthyol. Herpetol.):738.
- VINOGRADOV, A. P. 1953. Elementary composition of Pisces, Chapt. 21, pp. 463–566. In: *The Elementary Composition of Marine Organisms*. Yale Univ. Press, New Haven.
- WHITE, A. B., P. HANDLER, and E. L. SMITH. 1964. *Principles of Biochemistry*. McGraw-Hill, N. Y., p. 261.